

DOCS1-5976553-v1-P023389US\_Sequence\_listing  
SEQUENCE LISTING



<110> Hellenic Pasteur Institute  
Association Francaise Contre Les Myopathies  
Tzartos, Socrates  
Mamalaki, Avgi

<120> Production of recombinant fragments of muscle acetylcholine  
receptor and their use for ex vivo immunoadsorption of anti-Ch  
receptor antibodies from myasthenic patients

<130> P023389US

<140> US 10/512,004

<141> 2003-04-15

<150> GR 20020100190

<151> 2002-04-17

<160> 19

<170> PatentIn version 3.5

<210> 1

<211> 233

<212> PRT

<213> Pichia pastoris

<220>

<221> CARBOHYD

<222> (143)..(143)

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Glu Phe Ser Glu His Glu Thr Arg Leu Val Ala Lys Leu Phe Lys Asp  
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Tyr Ser Ser Val Val Arg Pro Val Glu Asp His Arg Gln Val Val Glu  
20 25 30

Val Thr Val Gly Leu Gln Leu Ile Gln Leu Ile Asn Val Asp Glu Val  
35 40 45

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Asn Gln Ile Val Thr Thr Asn Val Arg Leu Lys Gln Gln Trp Val Asp  
50 55 60

Tyr Asn Leu Lys Trp Asn Pro Asp Asp Tyr Gly Gly Val Lys Lys Ile  
65 70 75 80

His Ile Pro Ser Glu Lys Ile Trp Arg Pro Asp Leu Val Leu Tyr Asn  
85 90 95

Asn Ala Asp Gly Asp Phe Ala Ile Val Lys Phe Thr Lys Val Leu Leu  
100 105 110

Gln Tyr Thr Gly His Ile Thr Trp Thr Pro Pro Ala Ile Phe Lys Ser  
115 120 125

Tyr Cys Glu Ile Ile Val Thr His Phe Pro Phe Asp Glu Gln Asn Cys  
130 135 140

Ser Met Lys Leu Gly Thr Trp Thr Tyr Asp Gly Ser Val Val Ala Ile  
145 150 155 160

Asn Pro Glu Ser Asp Gln Pro Asp Leu Ser Asn Phe Met Glu Ser Gly  
165 170 175

Glu Trp Val Ile Lys Glu Ser Arg Gly Trp Lys His Ser Val Thr Tyr  
180 185 190

Ser Cys Cys Pro Asp Thr Pro Tyr Leu Asp Ile Thr Tyr His Phe Val  
195 200 205

Met Gln Arg Leu Glu Gln Lys Leu Ile Ser Glu Glu Asp Leu Asn Ser  
210 215 220

Ala Val Asp His His His His His His

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225

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<213> Pichia pastoris

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<222> (46)..(46)

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<222> (157)..(157)

&lt;400&gt; 2

Glu Ala Glu Ala Tyr Val Asp Tyr Lys Asp Asp Asp Asp Lys Glu Phe  
1 5 10 15

Arg Asn Gln Glu Glu Arg Leu Leu Ala Asp Leu Met Gln Asn Tyr Asp  
20 25 30

Pro Asn Leu Arg Pro Ala Glu Arg Asp Ser Asp Val Val Asn Val Ser  
35 40 45

Leu Lys Leu Thr Leu Thr Asn Leu Ile Ser Leu Asn Glu Arg Glu Glu  
50 55 60

Ala Leu Thr Thr Asn Val Trp Ile Glu Met Gln Trp Cys Asp Tyr Arg  
65 70 75 80

Leu Arg Trp Asp Pro Arg Asp Tyr Glu Gly Leu Trp Val Leu Arg Val  
85 90 95

Pro Ser Thr Met Val Trp Arg Pro Asp Ile Val Leu Glu Asn Asn Val  
100 105 110

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Asp Gly Val Phe Glu Val Ala Leu Tyr Cys Asn Val Leu Val Ser Pro  
115 120 125

Asp Gly Cys Ile Tyr Trp Leu Pro Pro Ala Ile Phe Arg Ser Ala Cys  
130 135 140

Ser Ile Ser Val Thr Tyr Phe Pro Phe Asp Trp Gln Asn Cys Ser Leu  
145 150 155 160

Ile Phe Gln Ser Gln Thr Tyr Ser Thr Asn Glu Ile Asp Leu Gln Leu  
165 170 175

Ser Gln Glu Asp Gly Gln Thr Ile Glu Trp Ile Phe Ile Asp Pro Glu  
180 185 190

Ala Phe Thr Glu Asn Gly Glu Trp Ala Ile Gln His Arg Pro Ala Lys  
195 200 205

Met Leu Leu Asp Pro Ala Ala Pro Ala Gln Glu Ala Gly His Gln Lys  
210 215 220

Val Val Phe Tyr Leu Leu Ile Gln Arg Lys His His His His His  
225 230 235 240

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<212> PRT  
<213> Pichia pastoris

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<222> (82)..(82)

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<222> (157)..(157)

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<400> 3

Glu Ala Glu Ala Tyr Val Asp Tyr Lys Asp Asp Asp Asp Lys Glu Phe  
1 5 10 15

Lys Asn Glu Glu Leu Arg Leu Tyr His His Leu Phe Asn Asn Tyr Asp  
20 25 30

Pro Gly Ser Arg Pro Val Arg Glu Pro Glu Asp Thr Val Thr Ile Ser  
35 40 45

Leu Lys Val Thr Leu Thr Asn Leu Ile Ser Leu Asn Glu Lys Glu Glu  
50 55 60

Thr Leu Thr Thr Ser Val Trp Ile Gly Ile Asp Trp Gln Asp Tyr Arg  
65 70 75 80

Leu Asn Tyr Ser Lys Asp Asp Phe Gly Gly Ile Glu Thr Leu Arg Val  
85 90 95

Pro Ser Glu Leu Val Trp Leu Pro Glu Ile Val Leu Glu Asn Asn Ile  
100 105 110

Asp Gly Gln Phe Gly Val Ala Tyr Asp Ala Asn Val Leu Val Tyr Glu  
115 120 125

Gly Gly Ser Val Thr Trp Leu Pro Pro Ala Ile Tyr Arg Ser Val Cys  
130 135 140

Ala Val Glu Val Thr Tyr Phe Pro Phe Asp Trp Gln Asn Cys Ser Leu  
145 150 155 160

Ile Phe Arg Ser Gln Thr Tyr Asn Ala Glu Glu Val Glu Phe Thr Phe  
165 170 175

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Ala Val Asp Asn Asp Gly Lys Thr Ile Asn Lys Ile Asp Ile Asp Thr  
180 185 190

Glu Ala Tyr Thr Glu Asn Gly Glu Trp Ala Ile Asp Phe Cys Pro Gly  
195 200 205

Val Ile Arg Arg His His Gly Gly Ala Thr Asp Gly Pro Gly Glu Thr  
210 215 220

Asp Val Ile Tyr Ser Leu Ile Ile Arg Arg Lys His His His His His  
225 230 235 240

His

<210> 4  
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<222> (159)..(159)

<400> 4

Glu Ala Glu Ala Tyr Val Asp Tyr Lys Asp Asp Asp Asp Lys Glu Phe  
1 5 10 15

Leu Asn Glu Glu Glu Arg Leu Ile Arg His Leu Phe Gln Glu Lys Gly

## DOCS1-5976553-v1-P023389US\_Sequence\_listing

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25

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Tyr Asn Lys Glu Leu Arg Pro Val Ala His Lys Glu Glu Ser Val Asp  
 35 40 45

Val Ala Leu Ala Leu Thr Leu Ser Asn Leu Ile Ser Leu Lys Glu Val  
 50 55 60

Glu Glu Thr Leu Thr Thr Asn Val Trp Ile Glu His Gly Trp Thr Asp  
 65 70 75 80

Asn Arg Leu Lys Trp Asn Ala Glu Glu Phe Gly Asn Ile Ser Val Leu  
 85 90 95

Arg Leu Pro Pro Asp Met Val Trp Leu Pro Glu Ile Val Leu Glu Asn  
 100 105 110

Asn Asn Asp Gly Ser Phe Gln Ile Ser Tyr Ser Cys Asn Val Leu Val  
 115 120 125

Tyr His Tyr Gly Phe Val Tyr Trp Leu Pro Pro Ala Ile Phe Arg Ser  
 130 135 140

Ser Cys Pro Ile Ser Val Thr Tyr Phe Pro Phe Asp Trp Gln Asn Cys  
 145 150 155 160

Ser Leu Lys Phe Ser Ser Leu Lys Tyr Thr Ala Lys Glu Ile Thr Leu  
 165 170 175

Ser Leu Lys Gln Asp Ala Lys Glu Asn Arg Thr Tyr Pro Val Glu Trp  
 180 185 190

Ile Ile Ile Asp Pro Glu Gly Phe Thr Glu Asn Gly Glu Trp Glu Ile  
 195 200 205

DOCS1-5976553-v1-P023389US\_Sequence\_listing

Val His Arg Pro Ala Arg Val Asn Val Asp Pro Arg Ala Pro Leu Asp  
210 215 220

Ser Pro Ser Arg Gln Asp Ile Thr Phe Tyr Leu Ile Ile Arg Arg Lys  
225 230 235 240

<210> 5  
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<212> PRT  
<213> Pichia pastoris

<220>  
<221> CARBOHYD  
<222> (157)..(157)

<400> 5

Glu Ala Glu Ala Tyr Val Asp Tyr Lys Asp Asp Asp Asp Lys Glu Phe  
1 5 10 15

Ser Glu Ala Glu Gly Arg Leu Arg Glu Lys Leu Phe Ser Gly Tyr Asp  
20 25 30

Ser Ser Val Arg Pro Ala Arg Glu Val Gly Asp Arg Val Arg Val Ser  
35 40 45

Val Gly Leu Ile Leu Ala Gln Leu Ile Ser Leu Asn Glu Lys Asp Glu  
50 55 60

Glu Met Ser Thr Lys Val Tyr Leu Asp Leu Glu Trp Thr Asp Tyr Arg  
65 70 75 80

Leu Ser Trp Asp Pro Ala Glu His Asp Gly Ile Asp Ser Leu Arg Ile  
85 90 95

Thr Ala Glu Ser Val Trp Leu Pro Asp Val Val Leu Leu Asn Asn Asn



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100

105

110

Asp Gly Asn Phe Asp Val Ala Leu Asp Ile Ser Val Val Val Ser Ser  
115 120 125

Asp Gly Ser Val Arg Trp Gln Pro Pro Gly Ile Tyr Arg Ser Ser Cys  
130 135 140

Ser Ile Gln Val Thr Tyr Phe Pro Phe Asp Trp Gln Asn Cys Thr Met  
145 150 155 160

Val Phe Ser Ser Tyr Ser Tyr Asp Ser Ser Glu Val Ser Leu Gln Thr  
165 170 175

Gly Leu Gly Pro Asp Gly Gln Gly His Gln Glu Ile His Ile His Glu  
180 185 190

Gly Thr Phe Ile Glu Asn Gly Gln Trp Glu Asn Ile His Lys Pro Ser  
195 200 205

Arg Leu Ile Gln Pro Pro Gly Asp Pro Arg Gly Gly Arg Glu Gly Gln  
210 215 220

Arg Gln Glu Val Ile Phe Tyr Leu Ile Ile Arg Arg Lys His His His  
225 230 235 240

His His His

<210> 6

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<212> DNA

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<223> Primer

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<400> 6  
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28

<210> 7  
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<223> Primer

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31

<210> 8  
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gcggaattct cggaggcgga gggctgac

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<210> 9  
<211> 54  
<212> DNA  
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<223> Primer

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<210> 10  
<211> 30  
<212> DNA  
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<220>

<223> Primer

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ggtgtagaat tccggaacca ggaggagcgc 30

<210> 11

<211> 56

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

<400> 11

atagtttagc ggccgcttag tgatggtgat ggtgatgctt gcgctggatg agcagg 56

<210> 12

<211> 28

<212> DNA

<213> Artificial Sequence

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<223> Primer

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ggtgtagaat tcaagaacga ggactgcg 28

<210> 13

<211> 59

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

<400> 13

atagtttagc ggccgcttag tgatggtgat ggtgatgctt ccggcggatg atgagcgag 59

<210> 14

<211> 8

<212> PRT

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<213> Artificial Sequence

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<223> FLAG amino acid sequence

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<223> Synthetic oligonucleotide

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gtagattaca aggatgacga tgacaaag 28

<210> 16

<211> 25

<212> DNA

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<223> Primer

<400> 16

gtgtggcagc gaattcctga acgag 25

<210> 17

<211> 25

<212> DNA

<213> Artificial Sequence

<220>

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gatgtagaat tctcacttgc ggcgg 25

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<210> 18

<211> 79

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

<400> 18

gcggccgcat gaaggttctg tgggctgcgt tgctggcac attcctggca ggatgccagg 60

cctccgaaca tgagacccg 79

<210> 19

<211> 40

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

<400> 19

ccgagcctcg agtcaatgat gatgatgatg atggtcgacg 40